



Individual capacity for repair of DNA damage and potential uses of stem cell lines for clinical applications: a matter of (genomic) integrity

Rumena Petkova, Nikolai Zhelev, Roumen Pankov & Stoyan Chakarov

To cite this article: Rumena Petkova, Nikolai Zhelev, Roumen Pankov & Stoyan Chakarov (2018): Individual capacity for repair of DNA damage and potential uses of stem cell lines for clinical applications: a matter of (genomic) integrity, *Biotechnology & Biotechnological Equipment*, DOI: [10.1080/13102818.2018.1520611](https://doi.org/10.1080/13102818.2018.1520611)

To link to this article: <https://doi.org/10.1080/13102818.2018.1520611>



© 2018 The Author(s). Published by Informa UK Limited, trading as Taylor & Francis Group.



Published online: 15 Oct 2018.



Submit your article to this journal [↗](#)



View Crossmark data [↗](#)

Individual capacity for repair of DNA damage and potential uses of stem cell lines for clinical applications: a matter of (genomic) integrity

Rumena Petkova^a, Nikolai Zhelev^b , Roumen Pankov^c  and Stoyan Chakarov^c 

^aFaculty of Medicine, Sofia University 'St. Kliment Ohridski', Sofia, Bulgaria; ^bCMCB, School of Science, Engineering & Technology, Abertay University, Dundee, UK; ^cDepartment of Biochemistry, Faculty of Biology, Sofia University 'St. Kliment Ohridski', Sofia, Bulgaria

ABSTRACT

Public and private human stem cell banking institutions are currently hosting hundreds of thousands partially characterized cell populations, including a significant number of human pluripotent stem cell lines. To be considered for use in clinical applications, stem cell preparations must undergo rigorous testing in order to ensure safety for the recipient. With development of the methodologies for in vitro derivation, ex vivo maintenance and expansion of stem cells and targeted differentiation of multipotent and pluripotent stem cells, many novel issues were added to the list of safety concerns of cell and tissue preparations. These issues are related to the potential changes that may occur in the course of in vitro propagation of stem cells and cell-derived products, how these changes may affect the quality of the preparation; and the potential effects on the recipient. Only a limited number of studies about the role of subtle variations of individual capacity for repair of genotoxic damage in maintenance in vitro of human stem cells are currently available. Nevertheless, the assessment of individual repair capacity may play a crucial role in the safety of use of human stem cells, as it constitutes a major factor in the risk of occurrence of genomic alterations that may seriously compromise the quality of the product. This article reviews the available data about the role of individual capacity for DNA damage repair in different human stem cell types and the potential adverse effects that may occur with the use of cell preparations with inferior repair capacity.

Abbreviations: AMD: age-related macular degeneration; DSB: double-strand break; ERCC2: excision repair cross-complementation group 2; ESC: embryonic stem cell; GVHD: graft-versus-host disease; HPSC: haematopoietic stem cell; iPSC: induced pluripotent stem cell; MSC: mesenchymal stem cell; NHEJ: non-homologous end joining; XPC: xeroderma pigmentosum complementation group C; XPD: xeroderma pigmentosum complementation group D; XRCC1: X-ray repair cross-complementing protein 1; XRCC3: X-ray repair cross-complementing protein 3

ARTICLE HISTORY

Received 19 June 2018

Accepted 4 September 2018





KEYWORDS

Aging; DNA repair; individual repair capacity; in vitro culturing; risk

Individual capacity for repair of genotoxic damage as a marker of the proliferation and differentiation capacity of stem cells

Cell therapy has become a legitimate therapeutic tool for haematological disease, inherited metabolic disorders, autoimmune disease, some types of solid tumours such as neuroblastoma, and, lately, for multiple sclerosis refractory to other types of treatment [1–5]. Safety for the recipient has always been a crucially important part of the criteria used to assess the potential of cell preparations for clinical applications. This includes not only concerns about the risk of potential transmission of infectious agents, but also short-term effects such as survival of transplanted cells

in the recipient, engraftment capability, capacity for rapid restoration of lost function and acute immunity-mediated effects such as acute graft-versus-host disease (GVHD); and long-term effects such as lifespan of the transplanted cells, self-renewing capacity of the stem cell niche, maintenance of production of differentiated cells, risk of recurrence of the primary disease (especially in autologous transplantations), risk of development of secondary disease (usually, neoplasia originating from the transplanted cells) and chronic immunity-mediated effects (chronic GVHD and others). With development of the methodologies for in vitro derivation, ex vivo maintenance and expansion of cells and targeted differentiation of multipotent and

CONTACT Stoyan Chakarov  stoianchakarov@gmail.com  Department of Biochemistry, Faculty of Biology, Sofia University 'St. Kliment Ohridski', Sofia, Bulgaria; Nikolai Zhelev  n.zhelev@abertay.ac.uk  CMCBR, School of Science, Engineering & Technology, Abertay University, Dundee, UK

© 2018 The Author(s). Published by Informa UK Limited, trading as Taylor & Francis Group.

This is an Open Access article distributed under the terms of the Creative Commons Attribution License (<http://creativecommons.org/licenses/by/4.0/>), which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited.

pluripotent stem cells, a whole host of other issues was added to the list of safety concerns of cell and tissue preparations. These issues are related to the potential changes that may occur in the course of in vitro propagation of stem cells and cell-derived products, how these changes may affect the quality of the preparation; and the potential effects on the recipient.

The proliferation potential of stem cells is typically higher than the potential of somatic cells (although the magnitude may significantly vary). Nevertheless, cultured stem cells may age and/or may be susceptible to carcinogenic transformation. The former may result in lower survival, lower engraftment rates, poor restoration of function and/or secondary disease (e.g. aplastic anemia originating from transplanted cells); whereas the latter may significantly increase the risk of secondary cancer in the recipient. Already, there have been reports about failure to sustain hematopoietic engraftment after transplantation of ex vivo expanded haematopoietic stem cells (HPSCs), development of secondary malignancies in patients with transplanted HPSCs and human pluripotent cell lines prone to genomic instability with potentially oncogenic genomic rearrangements after repeated passaging [6–9].

Detection and repair of genotoxic damage/maintenance of genomic integrity is critically important for progression through the cell cycle. For non-transformed somatic cells, cells that fail to comply with the requirements of the major genomic integrity checkpoints of the cell cycle (predominantly, the G1/S checkpoint) are forced into replicative senescence and/or apoptosis. Stem cells are inherently vulnerable to genotoxic damage for a variety of reasons. Among these, prominent are the ‘relaxed’ state of the chromatin and the fact that the main damage checkpoint in G1/S may not be functioning at its full efficiency in cells of early embryos (and, respectively, embryonic stem cells [ESCs]) [10,11]. Therefore, cells of early embryos and ESCs that have sustained damage may be routed to programmed cell death but may also be induced to differentiate, as differentiated cells typically have a functioning G1/S checkpoint [11–13]. The propensity of embryonic cells to sustain genomic damage is at least partly overcome by increased efficiency of the DNA repair machinery and activation of specific mechanisms for maintenance of genomic integrity in embryonic cells, such as *Filii*-dependent activation of Parp-1 [12,14,15]. There are, however, genetic factors that determine the individual parameters of the efficiency of DNA repair. There is some degree of inter-individual variance in the capacity to detect and repair DNA damage even among clinically healthy individuals

[16,17]. This variance may become significant with age (whether normal aging of the organism or aging of cultured cells), in conditions of increased genotoxic stress (e.g. after genotoxic treatments) and may be associated with adverse effects on the viability, proliferation and differentiation capacity of cells and cell products that may potentially be used for transplantation purposes. Several dozens of polymorphisms in key genes coding for products functioning in DNA repair/maintenance of genomic integrity have already been described (analyzed in detail in [17]). Alone or in combination with other factors, carriership of these polymorphisms may increase the risk of degenerative disease and/or cancer. When one or more of these polymorphisms are present in stem cell preparations intended for transplantation in human patients, it is possible that their otherwise subtle effects on the phenotype may become significant. It may be advisable to include molecular analysis of the capacity to detect and repair DNA damage in the panels for analysis of safety of cell and cell-derived preparations for transplantation purposes.

There have been a couple of studies about the role of efficiency of DNA repair for genomic stability in cultured lymphocytes showing that carriership of variant alleles of common polymorphisms in lymphocytes grown in vitro was associated with increased rates of chromosomal instability [18,19]. The amount of experimental data about the role of genetic polymorphisms of DNA repair genes in the risk of development of transplant-related complications is still limited.

Role of individual repair capacity for the outcomes of treatments based on HPSC

At present, the type of stem cells most commonly used for transplantation purposes are HPSCs and, respectively, the largest body of safety data pertains to HPSC transplantations. Transplantations of HPSCs are often preceded by ablation of the host haematopoiesis (myeloablative conditioning) and are followed by immunosuppressive treatments. Conditioning regimens typically include high-dose genotoxic agents that have severe adverse effects on tissues and organs other than the bone marrow, especially in tissues where cells have rapid natural turnover such as the skin and the gastrointestinal tract. The recovery may therefore be protracted and serious complications such as GVHD arise in no less than 35% of the recipients [20]. Acute GVHD results predominantly from the genotoxic damage inflicted by the pre-transplantation conditioning and its grade may be correlated with survival (typically, higher-

grade acute GVHD is associated with shorter survival). Chronic GVHD arises as a result of the attack of the immunocompetent cells in the graft on the recipient's tissues and may be associated with longer survival [21]. Subtle deficiencies in the capacity to manage genotoxic damage may be at least partly responsible for the risk of development of acute high-grade GVHD. At present, several markers for individual capacity for DNA repair (rs159153 in the *hOGG1* gene (coding for a glycosylase responsible for removal of oxidized bases from DNA); rs3135974 in the *LIG3* gene (coding for ligase III that is responsible for end joining in the late stages of repair by more than one mechanism); rs3219463 and rs3219476 in the *MUTYH* gene (coding for A/G mispair-specific adenine DNA glycosylase); rs6844176 in the *RFC-1* gene (coding for a component of the BRCA1-associated genome surveillance complex that scans DNA for the presence of damage) and the rs41376448 in the *HMGB1* gene (coding for HMGB1, a master regulator of transcription and DNA repair)) have been linked to the risk of acute and chronic GVHD and other causes of transplant-related mortality [22–24].

In patients with transplantations of HPSCs, there is always a risk of recurrence of the original tumour and for development of secondary malignancies, the risk being slightly higher in patients that have received autologous grafts [9]. There is already a report that grafts carrying variant alleles of genes coding for proteins of DNA repair (specifically, the rs1052559 polymorphism in the *XPD (ERCC2)* gene and rs861539 in the *XRCC3* gene) may be associated with higher risk of development of secondary leukemia [25].

Studies carried out in mouse models show that downregulation of the capacity for repair of double-strand breaks (DSBs) in DNA decreased the capacity for engraftment of HPSC [26–28]. The effect was observed with several key proteins of double-strand break repair (*Lig4*, *Ku80*, *Exo1*) and was noted to become significant with in vitro aging of the cells in the graft. This 'aging effect' is a hallmark of subtle genetic deficiencies of DNA repair/management of genomic integrity [17].

Role of individual repair capacity for research and potential clinical applications involving stem cells other than HPSC

The data about the role of individual capacity for repair of DNA/maintenance of genomic integrity for the outcomes of transplantations of stem cells other than HPSCs are still sparse. Presently, mesenchymal stem cells (MSCs) are viewed as another potential

source of stem cells for transplantation purposes. Bone marrow MSCs may be induced to differentiate to produce several different types of specialized cells, including osteocytes, chondrocytes, adipocytes and endothelial cells [29]. MSCs from Wharton's jelly of the umbilical cord and from placenta are believed to be pluripotent [30]. Multipotent bone marrow MSCs have been shown to be relatively resistant to DNA damage inflicted by inhibition of topoisomerases [31]. Studies using ex vivo expanded MSCs, however, showed that the efficiency of repair of double-strand breaks in DNA tends to decline after repeated passaging [32]. To date, freshly isolated or expanded ex vivo autologous MSCs from bone marrow have been used in experimental therapy of human patients with stroke and spinal cord injury [33]. Assessment of long-term adverse effects was carried out for the latter study and was reported as negative [34]. Nevertheless, the number of in vivo studies of safety of transplantation of MSCs does not permit reliable assessment of the potential risks. hESCs have been (and still are) a 'gold standard' in studies using human pluripotent cells but increased use of human induced pluripotent stem cells (iPSCs) lines has been recently reported [35]. According to the Human Pluripotent Stem Cell Registry (https://hpscereg.eu/news/single_news?id=72; retrieved 2018 Apr 11), nearly 2500 human pluripotent stem cell lines (over 700 hESC lines and about 1800 iPSC lines) are presently listed as globally available. iPSCs have been intensely studied with regard to their potential applications for the reparative and regenerative medicine. Their use was initially limited to research purposes only for a variety of reasons, including 'incomplete' reprogramming (iPSC continue to express genes typical of the cell type that they were derived from); 'cancer-like' expression profile, accelerated aging, low survival and lower efficiency of differentiation [36–38]. Recently, potential future clinical applications have begun to emerge for iPSCs as well as ESCs, including age-related macular degeneration (AMD), vascular disease, diabetes and Parkinson's disease [39–42]. It was demonstrated that human iPSCs deficient for the *LIG4* gene (coding for the main ligase of repair of DSBs by non-homologous end joining (NHEJ)) exhibited significant decrease in reprogramming efficiency than iPSC derived from normal human cells and rapidly accumulated chromosomal abnormalities [43]. Targeted differentiation of these *LIG4*-deficient iPSCs into haematopoietic progenitors was impaired, resulting in accumulation of DSBs and high rates of apoptosis. Later, it was shown that iPSCs without apparent genetic defects subjected to prolonged in vitro passaging

accumulated DSBs and exhibited mitochondrial dysfunction, increased rates of apoptosis and decreased differentiation efficiency [44,45]. Apparently, DNA repair is an important factor for maintenance of stemness qualities of iPSC. iPSCs derived by more recent modified methods were reported to resemble ESCs more than iPSCs derived by traditional methods [46]. Among the main differences were increased capacity for repair of double-strand breaks and increased accuracy of NHEJ for iPSCs derived by improved methodology, therefore, the safety of iPSC preparations in terms of DNA repair capacity has apparently been improved. Nevertheless, other safety concerns might arise. These are related to the fact that there is always an inherent risk that iPSCs derived from the patient's own cells may exhibit, in the course of their establishment or in vitro propagation, a propensity for genetic instability. Since carriage of polymorphisms in genes coding for proteins directly or indirectly responsible for the maintenance of genomic integrity is not uncommon, this risk may be significant when using autologous cells. Already, there has been one suspended trial of treatment of human disease with iPSCs after discovering that the autologous iPSCs intended to be used for treatment of AMD has sustained potentially dangerous mutations [47]. Consequently, designated acts regarding the safety of stem cell preparations have been introduced (the Act on the Safety of Regenerative Medicine (2014) in Japan, where the trial was conducted, and the 21st Century Cures Act in the USA (2016). The trials were only recently resumed after the project leader Masayo Takahashi and the Nobel laureate for 2012 Shinya Yamanaka declared that the cells used further in the project will be provided by the cell bank of Kyoto University's Center for iPSC Cell Research and Application (headed by Yamanaka) in order to guarantee that they have been comprehensively characterized and tested for quality. A list of recommendations regarding future trials using iPSCs have been made, including specific recommendations for the assessment of potential risks; the choices and decisions of the participants free of therapeutic misconception and individualized care for the patients participating in the trial [48]. Nevertheless, the eye is a relatively immunoprivileged site. Potential uses of autologous iPSCs for derivation of differentiated cells and tissues for other than ophthalmological purposes may be limited by histocompatibility issues. Recently, several clinical trials for potential uses of iPSCs in neurological disease have been launched [49,50] but no results have been reported yet. Assessment of the individual capacity for

DNA repair/maintenance of genomic integrity may assist in the screening of potential patients to differentiate between patients that may be eligible for transplantations of autologous iPSC-derived cells and patients that may benefit from cell preparations from allogeneic iPSCs.

Reports about the role of the capacity for repair of DNA damage/maintenance of genomic integrity in ESCs are still quite rare in the specialized literature. The intact functioning of the mechanism for repair of DNA is of prime importance for ESCs, as their restriction point of the cell cycle is weak (in human ESCs) or virtually non-existent (in murine ESCs) [11–13]. Repair of DSBs in ESCs is important, as with all types of stem cells. This may be especially valid for repair by homologous recombination, as it was shown to be used as a preferred repair pathway throughout the cell cycle, at least in murine ESCs [51]. It has been recently demonstrated that the XPC-HR23B complex (normally responsible for the identification of genomic damage in untranscribed regions) may play a role in the control of the 'stemness' state in human hESCs [52,53]. This was implemented by activation of transcription of OCT4 and SOX2 and by regulation of demethylation of DNA via rapid excision-synthesis-end ligation cycle that generated methylation-free DNA regions faster than the 'conventional' thymine-DNA glycosylase-mediated base excision repair. Differentiation of ESCs into specialized cell types may be dependent on the efficiency of specific types of DNA repair. Specifically, differentiation along the myogenic line in human ESCs requires temporal stimulation of the DNA repair mediated by XRCC1 (a stabilizing factor of DNA ligase III) [54].

Only recently, it has been reported that China was on the verge of launching the first ever clinical trial for use of hESC-derived cell products in treatment of Parkinson's disease [55]. It is still too early for any results, but nevertheless, it could be expected that the potential safety concerns would be, at least partly, related to genomic change occurring in the process of cell maintenance in vitro.

Apparently, the efficiency of repair of DNA damage is an important factor for maintenance of stem cells in culture, for efficient reprogramming back to pluripotent state and for differentiation into specific cell types. It could be expected that subtle deficiencies of DNA repair/maintenance of genomic integrity may affect the survival of stem cells in culture, especially with increased passaging. Moreover, even for the stem cell preparations prepared in strict xeno-free conditions, the contact with agents with potentially

genotoxic effects such as DMSO (dimethyl sulphoxide) is, at present, unavoidable. Only a limited number of studies about the role of subtle variations of individual repair capacity in the maintenance of stem cell lines in vitro are currently available [56,57]. Since the applications of stem cells and stem cell-derived products continue to expand and diversify, it might be advisable to augment the currently existing panel of markers used for characterization of stem cells and stem cell lines with markers for individual repair capacity in order to improve the process of selection for lines with potential use in clinical applications.

Conclusions

Knowledge about the individual DNA repair capacity of human stem cell lines may be valuable for the purposes of research, especially in light of potential genotype–phenotype correlations. Use of cells and cell lines with capacity to repair genotoxic damage that is inferior to the average may be undesirable for potential clinical applications because of higher risks of engraftment failure and/or potential long-term complications. Analysis of individual capacity for repair of genotoxic damage/maintenance of genomic integrity may become a tool in the assessment of the capacity for proliferation and differentiation of human stem cell lines with regard to their potential long-term maintenance in vitro and the potential in vivo applications.

Disclosure Statement

The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

Funding

This research was supported by the National Science Fund, Ministry of Education and Science of Republic of Bulgaria under grant number DFNI-B01/2.

ORCID

Nikolai Zhelev  <http://orcid.org/0000-0002-5189-3975>
 Roumen Pankov  <http://orcid.org/0000-0002-3157-365>
 Stoyan Chakarov  <http://orcid.org/0000-0002-0712-9793>

References

- [1] Yalçın B, Kremer LC, van Dalen EC. High-dose chemotherapy and autologous haematopoietic stem cell rescue for children with high-risk neuroblastoma. *Cochrane Database Syst Rev*. 2015;(10):CD006301. DOI: [10.1002/14651858.CD006301.pub3](https://doi.org/10.1002/14651858.CD006301.pub3)
- [2] Chivu-Economescu M, Rubach M. Hematopoietic Stem Cells Therapies. *Curr Stem Cell Res Ther*. 2017;12(2): 124–133.
- [3] Muraro PA, Martin R, Mancardi GL, et al. Autologous haematopoietic stem cell transplantation for treatment of multiple sclerosis. *Nat Rev Neurol*. 2017;13(7): 391–405.
- [4] Penati R, Fumagalli F, Calbi V, et al. Gene therapy for lysosomal storage disorders: recent advances for metachromatic leukodystrophy and mucopolysaccharidosis I. *J Inherit Metab Dis*. 2017;40(4):543–554.
- [5] Zeher M, Papp G, Nakken B, et al. Hematopoietic stem cell transplantation in autoimmune disorders: From immune-regulatory processes to clinical implications. *Autoimmun Rev*. 2017;16(8):817–825.
- [6] Holyoake TL, Alcorn MJ, Richmond L, et al. CD34 positive PBPC expanded ex vivo may not provide durable engraftment following myeloablative chemoradiotherapy regimens. *Bone Marrow Transplant*. 1997;19(11): 1095–1101.
- [7] Draper JS, Smith K, Gokhale P, et al. Recurrent gain of chromosomes 17q and 12 in cultured human embryonic stem cells. *Nat Biotechnol*. 2004;22:53–54.
- [8] Hovatta O, Jaconi M, Töhönen V, et al. A teratocarcinoma-like human embryonic stem cell (hESC) line and four hESC lines reveal potentially oncogenic genomic changes. *PLoS One*. 2010;5(4):e10263. DOI:[10.1371/journal.pone.0010263](https://doi.org/10.1371/journal.pone.0010263)
- [9] Vaxman I, Ram R, Gaftor-Gvili A, et al. Secondary malignancies following high dose therapy and autologous hematopoietic cell transplantation-systematic review and meta-analysis. *Bone Marrow Transplant*. 2015;50(5):706–714.
- [10] Becker KA, Ghule PN, Therrien JA, et al. Self-renewal of human embryonic stem cells is supported by a shortened G1 cell cycle phase. *J Cell Physiol*. 2006; 209(3):883–893.
- [11] Bárta T, Vinarský V, Holubcová Z, et al. Human embryonic stem cells are capable of executing G1/S checkpoint activation. *Stem Cells*. 2010;28(7):1143–1145.
- [12] Arabadjev A, Petkova R, Momchilova A, et al. Of mice and men – differential mechanisms of maintaining the undifferentiated state in mESC and hESC. *BioDiscovery*. 2012;3:e8927. DOI:[10.7750/BioDiscovery.2012.3.1](https://doi.org/10.7750/BioDiscovery.2012.3.1)
- [13] Laugesen A, Helin K. Chromatin repressive complexes in stem cells, development, and cancer. *Cell Stem Cell*. 2014; 14(6):735–751.
- [14] Nagaria P, Robert C, Rassool FV. DNA double-strand break response in stem cells: mechanisms to maintain genomic integrity. *Biochim Biophys Acta*. 2013; 1830(2):2345–2353.
- [15] Zhao B, Zhang WD, Duan YL, et al. Filia is an ESC-specific regulator of DNA damage response and safeguards genomic stability. *Cell Stem Cell*. 2015;16(6): 684–698.
- [16] Pero RW, Bryngelsson C, Mitelman F, et al. Interindividual variation in the responses of cultured human lymphocytes to exposure from DNA damaging chemical agents: interindividual variation to carcinogen exposure. *Mutat Res*. 1978;53(3):327–341.

- [17] Chakarov S, Petkova R, Russev GCh. Individual capacity for detoxification of genotoxic compounds and repair of DNA damage. Commonly used methods for assessment of capacity for DNA repair. *BioDiscovery*. 2014;11:e8958. DOI:[10.7750/BioDiscovery.2014.11.2](https://doi.org/10.7750/BioDiscovery.2014.11.2)
- [18] Vodicka P, Kumar R, Stetina R, et al. Genetic polymorphisms in DNA repair genes and possible links with DNA repair rates, chromosomal aberrations and single-strand breaks in DNA. *Carcinogenesis*. 2004;25(5):757–763.
- [19] Shkarupa VM, Mishcheniuk OY. Polymorphism of DNA repair gene XPD Lys751Gln and chromosome aberrations in lymphocytes of thyroid cancer patients exposed to ionizing radiation due to the Chernobyl accident. *Exp Oncol*. 2016;38(4):257–260.
- [20] Jacobsohn DA, Vogelsang GB. Acute graft versus host disease. *Orphanet J Rare Dis*. 2007;2:35. DOI: [10.1186/1750-1172-2](https://doi.org/10.1186/1750-1172-2)
- [21] Valcárcel D, Martino R, Caballero D, et al. Sustained remissions of high-risk acute myeloid leukemia and myelodysplastic syndrome after reduced-intensity conditioning allogeneic hematopoietic transplantation: chronic graft-versus-host disease is the strongest factor improving survival. *J Clin Oncol*. 2008;26(4):577–584.
- [22] Arora M, Lindgren B, Basu S, et al. Polymorphisms in the base excision repair pathway and graft-versus-host disease. *Leukemia*. 2010;24:1470–1475.
- [23] Kornblit B, Mamas T, Petersen SL, et al. Association of HMGB1 polymorphisms with outcome after allogeneic hematopoietic cell transplantation. *Biol Blood Marrow Transplant*. 2010;16:239–252.
- [24] Thyagarajan B, Lindgren B, Basu S, et al. Association between genetic variants in the base excision repair pathway and outcomes after hematopoietic cell transplantations. *Biol Blood Marrow Transplant*. 2010;16:1084–1089.
- [25] Diamond HR, Ornellas MH, Orfao A, et al. Acute myeloid leukemia of donor origin after allogeneic stem cell transplantation from a sibling who harbors germline XPD and XRCC3 homozygous polymorphisms. *J Hematol Oncol*. 2011;4:39. DOI:[10.1186/1756-8722-4-39](https://doi.org/10.1186/1756-8722-4-39)
- [26] Nijnik A, Woodbine L, Marchetti C, et al. ANA DNA repair is limiting for haematopoietic stem cells during ageing. *Nature*. 2007;447(7145):686–690.
- [27] Niedernhofer LJ. DNA repair is crucial for maintaining hematopoietic stem cell function. *DNA Repair (Amst)*. 2008;7(3):523–529.
- [28] Desai A, Qing Y, Gerson SL. Exonuclease 1 is a critical mediator of survival during DNA double strand break repair in nonquiescent hematopoietic stem and progenitor cells. *Stem Cells*. 2014;32(2):582–593.
- [29] Abdallah BM, Kassem M. Human mesenchymal stem cells: from basic biology to clinical applications. *Gene Ther*. 2008;15(2):109–116.
- [30] Perin L, Sedrakyan S, Da Sacco S, et al. Characterization of human amniotic fluid stem cells and their pluripotential capability. *Methods Cell Biol*. 2008;86:85–99.
- [31] Nicolay NH, Rühle A, Perez RL, et al. Mesenchymal stem cells exhibit resistance to topoisomerase inhibition. *Cancer Lett*. 2016;374(1):75–84.
- [32] Hare I, Gencheva M, Evans R, et al. In vitro expansion of bone marrow derived mesenchymal stem cells alters DNA double strand break repair of etoposide induced DNA damage. *Stem Cells Int*. 2016;2016:8270464. DOI:[10.1155/2016/8270464](https://doi.org/10.1155/2016/8270464)
- [33] Pal R, Venkataramana NK, Bansal A, et al. Ex vivo-expanded autologous bone marrow-derived mesenchymal stromal cells in human spinal cord injury/paraplegia: a pilot clinical study. *Cytotherapy*. 2009;11(7):897–911.
- [34] Lee JS, Hong JM, Moon GJ, et al. A long-term follow-up study of intravenous autologous mesenchymal stem cell transplantation in patients with ischemic stroke. *Stem Cells*. 2010;28(6):1099–1106.
- [35] Kobold S, Guhr A, Kurtz A, et al. Human embryonic and induced pluripotent stem cell research trends: complementation and diversification of the field. *Stem Cell Rep*. 2015;4(5):914–925.
- [36] Chin MH, Mason MJ, Xie W, et al. Induced pluripotent stem cells and embryonic stem cells are distinguished by gene expression signatures. *Cell Stem Cell*. 2009;5(1):111–123.
- [37] Feng Q, Lu SJ, Klimanskaya I, et al. Hemangioblastic derivatives from human induced pluripotent stem cells exhibit limited expansion and early senescence. *Stem Cells*. 2010;28(4):704–712.
- [38] Ghosh Z, Wilson KD, Wu Y, Hu S, et al. Persistent donor cell gene expression among human induced pluripotent stem cells contributes to differences with human embryonic stem cells. *PLoS One*. 2010;5(2):e8975. DOI:[10.1371/journal.pone.0008975](https://doi.org/10.1371/journal.pone.0008975)
- [39] Doi D, Samata B, Katsukawa M, et al. Isolation of human induced pluripotent stem cell-derived dopaminergic progenitors by cell sorting for successful transplantation. *Stem Cell Rep*. 2014;2(3):337–350.
- [40] Schwartz SD, Regillo CD, Lam BL, et al. Human embryonic stem cell-derived retinal pigment epithelium in patients with age-related macular degeneration and Stargardt's macular dystrophy: follow-up of two open-label phase 1/2 studies. *Lancet*. 2015;385(9967):509–516.
- [41] Menasché P, Vanneaux V, Fabreguettes JR, et al. Towards a clinical use of human embryonic stem cell-derived cardiac progenitors: a translational experience. *Eur Heart J*. 2015;36(12):743–750.
- [42] Yabe SG, Fukuda S, Takeda F, et al. Efficient generation of functional pancreatic β -cells from human induced pluripotent stem cells. *J Diabetes*. 2017;9(2):168–179.
- [43] Tilgner K, Neganova I, Moreno-Gimeno I, et al. A human iPSC model of Ligase IV deficiency reveals an important role for NHEJ-mediated-DSB repair in the survival and genomic stability of induced pluripotent stem cells and emerging haematopoietic progenitors. *Cell Death Differ*. 2013;20(8):1089–1100.
- [44] Masotti A, Celluzzi A, Petrini S, et al. Aged iPSCs display an uncommon mitochondrial appearance and fail to undergo in vitro neurogenesis. *Aging (Albany NY)*. 2014;6(12):1094–1108.

- [45] Simara P, Tesarova L, Rehakova D, et al. DNA double-strand breaks in human induced pluripotent stem cell reprogramming and long-term in vitro culturing. *Stem Cell Res Ther.* 2017;8:73. DOI:[10.1186/s13287-017-0522-5](https://doi.org/10.1186/s13287-017-0522-5)
- [46] Nagaria PK, Robert C, Park TS, et al. High-fidelity reprogrammed human iPSCs have a high efficacy of DNA repair and resemble hESCs in their MYC transcriptional signature. *Stem Cells Int.* 2016;2016:3826249. DOI:[10.1155/2016/3826249](https://doi.org/10.1155/2016/3826249)
- [47] Garber K. RIKEN suspends first clinical trial involving induced pluripotent stem cells. *Nat Biotechnol.* 2015;33(9):890–891.
- [48] Takashima K, Inoue Y, Tashiro S, et al. Lessons for reviewing clinical trials using induced pluripotent stem cells: examining the case of a first-in-human trial for age-related macular degeneration. *Regen Med.* 2018;13(2):123–128.
- [49] Gazdic M, Volarevic V, Harrell CR, et al. Stem cells therapy for spinal cord injury. *Int J Mol Sci.* 2018;19(4):1039. DOI: [10.3390/ijms19041039](https://doi.org/10.3390/ijms19041039)
- [50] Takahashi J. Stem cells and regenerative medicine for neural repair. *Curr Opin Biotechnol.* 2018;52:102–108.
- [51] Serrano L, Liang L, Chang Y, et al. Homologous recombination conserves DNA sequence integrity throughout the cell cycle in embryonic stem cells. *Stem Cells Dev.* 2011;20(2):363–374.
- [52] Cattoglio C, Zhang ET, Grubisic I, et al. Functional and mechanistic studies of XPC DNA-repair complex as transcriptional coactivator in embryonic stem cells. *Proc Natl Acad Sci USA.* 2015;112(18):E2317–2326.
- [53] Ho JJ, Cattoglio C, McSwiggen DT, et al. Regulation of DNA demethylation by the XPC DNA repair complex in somatic and pluripotent stem cells. *Genes Dev.* 2017;31(8):830–844.
- [54] Al-Khalaf MH, Blake LE, Larsen BD, et al. Temporal activation of XRCC1-mediated DNA repair is essential for muscle differentiation. *Cell Discov.* 2016;2:15041. DOI:[10.1038/celldisc.2015.41](https://doi.org/10.1038/celldisc.2015.41)
- [55] Cyranoski D. Trials of embryonic stem cells to launch in China. *Nature.* 2017;546(7656):15–16.
- [56] Hanson C, Caisander G. Human embryonic stem cells and chromosome stability. *APMIS.* 2005;113(11-12):751–755.
- [57] Reynolds L. The success of stem cell transplantations and the potential post-transplantation complications may be dependent, among other factors, on the capacity of the recipient and the transplanted cells to repair DNA damage. *BioDiscovery.* 2016;19:e9076. DOI:[10.3897/BioDiscovery.19.e9076](https://doi.org/10.3897/BioDiscovery.19.e9076)